

The Preparation and Use of Artificial Diets for Rearing Insects

(Concluded from p. 270)

IGNOFFO'S MEDIUM

Developed by Dr. Carlo M. Ignoffo in Texas, U.S.A., this medium is a development of an earlier medium devised for feeding the Pink bollworm by Erma S. Vanderzant and Raymond Reiser. It is more sophisticated in many ways than Shorey's medium and lends itself to various modifications, in particular the varying of the "dried leaf powder" to suit different species. So far the following have been reared:— Pink bollworm (*Pectinophora gossypiellae*); Ni moth (*Plusia ni*); Cabbage moth (*Mamestra brassicae*); Large yellow underwing (*Triphaena pronuba*); Bufftip (*Phalera bucephala*); Garden tiger (*Artia caja*); Scarlet tiger (*Panaxia dominula*); Eri silkmoth (*Philosamia ricini*); Large, Small and Green-veined whites (*Pieris brassicae*, *P. rapae* and *P. napi*); Brazillian bullseye (*Automeris aurantiaca*); Blowfly (*Calliphora erythrocephala*); Housefly (*Musca domestica*); Fruitfly (*Drosophila melanogaster*) and Desert Locust (*Schistocerca gregaria*).

Partial success has also been obtained with *Automeris pyrromelas* and *Copaxa multifenestrata*, whose natural food-plant is unknown. Both species reached the 3rd or 4th instar but then expired of a virus disease. But that they feed at all is interesting and encouraging.

At 29°C. Ni moth larvae feed up in eight days on this medium. At lower temperatures and for longer life-cycles, the jars of medium must be changed at weekly intervals for fresh. At 20°C. Cabbage butterfly larvae take three weeks and require two changes of diet. This medium can be stored indefinitely in a deep-freeze. It should not be used after more than 7-10 days storage at 0°C., and then only for larger larvae.

The diet consists of the following ingredients:— Water, 220 ml.; Casein (light white soluble), 25.2 g.; Bemax, 21.6 g.; Sugar, 25.2 g.; Dried Leaf powder (may be omitted), 10.8 g.; Wessons salts, 7.2 g.; Cellulose powder CF11 (Whatman "Chromedia"), 3.6 g.; Choline chloride (10% Aqueous soln.), 7.2 ml.; Methy 4-hydroxy benzoate (15% Alcoholic soln.), 7.2 ml.; Vitamin soln., 1.2 ml.; Formalin (10%), 3 ml.; Mazola corn oil, 2 ml.

To prepare the diet mix all the above together using a good mixer, preferably a blender. The solid ingredients first to the water and then adding the liquid ingredients one at a time. A point to note is that if dried leaf powder is being used it is not then necessary to use the Mazola corn oil as well (see below). For *Pieris* species, 3.0 ml. of a 1% aqueous solution of sinigrin (Potassium myronate) may be substituted for dried cabbage leaf powder.

While the above is being mixed, boil the following

* A.R.C. Unit of Invertebrate Chemistry and Physiology, Department of Zoology, Downing Street, Cambridge.

together: — Water, 400 ml.; Agar, 18 g.

When boiled, allow to cool to 70°C. then mix into the other ingredients. Finally add: — Vitamin C (Ascorbic acid), 3.0 g.; Aureomycin (Veterinary grade), 1.6 g. Mix well in and pour immediately into the desired containers.

I have already mentioned that the dried leaf powder may be varied according to species. It may also be omitted altogether. Nevertheless, it does contain some vital ingredient and when, omitted, 2.0 ml. of "Mazola" corn oil *must be added in lieu*. Medium without dried leaf powder, but with oil, is entirely satisfactory, provided the larvae will start to feed. With normally polyphytophagous species such as Cabbage moth or Garden tiger, little difficulty is experienced, but with more specific feeders, such as the Crucifer feeding Large cabbage white, the initial "take" of newly-hatched larvae falls from 100% to 60%. It can be restored to 100% by adding to the medium a feeding stimulant, normally present in Crucifers, such as the mustard oil glucoside Sinigrin, to the extent of one part in 100,000. There remains great scope for individual experimentation in this field.

The jars of medium should be covered with filter paper or a "Kleenex" tissue, held on with a rubber band. They should be kept on their sides. This ensures that frass falls clear of the medium. It is best to keep the medium at 20°-25°C., and it should not be kept below 60% R.H. If too dry, the medium will rapidly dry out; a sure sign of this is an obvious shrinkage of the medium away from the sides of the jars. Should this occur the larvae must be transferred to a fresh batch.

At weekly intervals they should in any case be transferred. (Do not do this when they are sitting moulting! — wait until they have finished.) When the larvae reach the final instar, the tops should be replaced with terylene gauze to allow more ventilation. It now also becomes necessary to clean out the frass at least every other day if possible.

YAMAMOTO DIET

This diet was specifically designed for the rearing of Tobacco hornworm (*Manduca sexta*). It is usually stored at 0°C. when it keeps for 2-3 weeks. At -25°C. there is a tendency for the physical structure of the gel to break down, but the factors involved are complex and could appear to depend on the particular batch of agar used and also the final temperature of the completed mix.

This diet can be used without changing, for species with a larval life-cycle not exceeding three weeks. Again, this facility must be used with circumspection, as it depends on the number of larvae present and the rate of drying out. Unlike Ignoffo's mix, dried leaf powder is not to be added to this diet. It seems likely that yeast, which contains many things, may act as a general stimulant for a number of species (eg. *P. brassicae*) which will feed 100% on this diet without requiring the specific addition of any feeding stimulants. It has also

been suggested that the addition of the vitamin mixture may be unnecessary. I have not myself tested this hypothesis, but in view of the results obtained with Bot's diet (see below), this may well be so.

The ingredients are as follows:— Agar, 15 g.; Distilled water, 375 ml.; Casein, 26.25 g.; Bemax, 56.25 g.; Sugar, 22.50 g.; Dried yeast, 11.25 g.; Wessons salts, 7.50 g.; Sorbic acid, 1.12 g.; Cholesterol, 0.75g.; Methyl-4-benzoate, 0.75 g.; Choline chloride, 0.75 g.; Distilled water, 262.5 ml.; 10% Formaldehyde, 3.37 ml.; Corn oil, 1.50 ml.; Vitamin mixture, 1.8 ml.; Ascorbic acid, 3.00 g.; Aureomycin, 1.50 g.

This diet should be made as Ignoffo's, but the boiled agar only requires to be cooled to 90°C. prior to mixing in.

BOT'S DIET

This diet is a variation on the above, omitting some of the ingredients. It has been used successfully for several species of armyworm, some of which are difficult to rear on their normal foodplants. It has the advantage of not using the vitamin mixture. As originally described by Bot, the preparation is tedious and all the ingredients were finally heated to 90°C. (which could largely destroy the Vitamin C), and I would recommend the diet in fact to be made up in the same manner as Ignoffo's diet.

The ingredients are:— Casein, 8.0 g.; Bemax, 52.0 g.; Yeast, 50.0 g.; Choline chloride, 0.4 g.; Cholesterol, 0.2 g.; Inositol, 0.2 g.; Methyl-4-benzoate, 2.6 g.; Ascorbic acid, 5.0 g.; Agar, 12.0 g.; Water, 630 ml.

Bot's original instructions for this diet are as follows:— To prepare the medium, the wheat germ, caesin, and yeast were thoroughly mixed. Methyl p-hydroxbenzoate and cholesterol were dissolved in 20 ml. of 95% ethanol and stirred into the above mixture, taking care to moisten the total contents evenly. The alcohol was then evaporated, either by constant stirring of the mixture in a steam bath, or by heating to 60°C. under vacuum, or simply by spreading out the mixture in a thin layer in a warm place and allowing it to dry. The dry mixture was allowed to return to room temperature, and then the agar was thoroughly mixed into it. Inositol, choline and ascorbic acid were dissolved in the water, and then stirred into the mixture. The resulting thin gruel was poured into 4 x 1 in. specimen tubes, to a depth of about $\frac{3}{4}$ in. These were then stoppered with cottonwool, and placed in a slanting position in an oven at 90°C. for half an hour until a soft, light brown crust, has formed on the medium. The tubes were allowed to cool in an upright position. When water condensed in the tubes, they were set aside for 24 hours to allow the moisture to return to the medium.

Discussion

These artificial diets have all been developed to supply a need. Nearly all the work has been done in the U.S.A., and apart from some Research Institutes, they are as yet little

known in England. They deserve to be far better known. They can be of great benefit not only to the professional, but also to the Amateur Entomologist. Their preparation is comparatively easy. It has been shown in at least one instance, that mortality is invariably less than when on a natural or substitute foodplant. The number of diet formulations now published must run into several hundreds. Many of these are variations on the same theme, with but slight variation and perhaps designed for different species. It does indeed seem that quite large liberties can be taken with varying, omitting, and adding to the ingredients; the field is in fact wide open. Table III gives a summary of various species which have been reared on the various diets, either by the original author of the diet formulation, or by the present author. Apart from the original references to the diets, attention should be given to two recipe books by House (1967), and House, Singh and Balsch (1971), which are a mine of information on the various diets which have been formulated for use with various insect species.

TABLE III

Species which have been successfully reared on the four diets described.

SHOREY'S

Trichoplusia ni; *Autoplusia egea*; *Autographa californica*; *Heliothis phloxiphaga*; *Spodoptera exigua*; *Pseudaletia unipuncta*; *Prodenia ornithogalli*; *Peridroma saucia*; *Heliothis zea*; *Mamestra brassicae*.

IGNOFFO'S

Trichoplusia ni; *Philosamia ricini*; *Pieris brassicae*; *Mamestra brassicae*; *Pieris rapae*; *Manduca sexta*; *Pieris napi*; *Carausius morosus*; *Arctia caja*; *Schistocerca gregaria*; *Lymantra dispar*; *Calliphora ethrocephala*; *Triphoena pronuba*; *Drosophila melanogaster*; *Papilio machaon*; *Musca domestica*.

YAMAMOTO'S

Manduca sexta; *Trichoplusia ni*; *Manduca quinque-maculata*; *Schistocerca gregaria*; *Pieris brassicae*; *Calliphora ethrocephala*; *Arctia caja*; *Drosophila melanogaster*; *Lymantra dispar*, *Musca domestica*; *Phlogophora meticulosa*; *Spodoptera exempta*; *Danaus plexippus*.

BOT'S

Spodoptera exempta; *Heliothis armigera*; *Spodoptera exigua*; *Prodenia litura*; *Spodoptera cilium*; *Plusia acuta*.

References

- Bot, J., 1967. An artificial rearing medium for three Noctuids of economic importance belonging to the genus *Spodoptera* (Lepidoptera). *J. ent. Soc. S. Afr.*, 29: 157-160.

- House, H. L., 1967. Artificial diets for insects: A compilation of references with abstracts. *Info. Bull. Res. Inst. Can. Dept. Agric.*, No. 5: 1-163.
- House, H. L., Singh, P., and Batsch, W. W., 1971. Artificial diets for insects: A compilation of references and abstracts. *Info. Bull. Res. Inst. Can. Dept. Agric.*, No. 7: 1-156
- Ignoffo, C. M., 1963. A successful technique for mass rearing of cabbage loopers on a semisynthetic diet. *Ann. Entomol. Soc. Amer.*, 56: 178-182.
- Shorey, H. H. and Hale, R. L., 1965. Mass-rearing of the larvae of nine noctuid species on a simple artificial medium. *J. econ. Ent.*, 58: 522-524.
- Yamamoto, R. T., 1969. Mass rearing of the tobacco hornworm, II. Larval rearing and pupation. *J. econ. Ent.*, 62: 1427-1431.

ADDENDUM

Since this paper was written the basic diet ingredients have become available in England through ICN Pharmaceuticals Inc., Riverdale Estate, Molesay Road, Hersham, Surrey, and Plenum Publishing Corporation have published a very comprehensive, but expensive, receipt book by Pritam Singh entitled "Artificial diets for Insects, Mites and Spiders", price £47.50.

TRICHIUS FASCIATUS (L.) (COL.: SCARABAEIDAE) IN NORTH WALES. — While walking in the Coed y Brenin Forest, north of Dolgellau, on the 16th July, 1978, a strange looking Bumblebee was seen to enter a Foxglove flower. On capture it proved to be the Bee-beetle, *Trichius fasciatus* (Linn.), a well-known 'mimic' of the Bumblebee. The only other record for Merionethshire I can find for this insect, described as rare in Wales, is in the Llanbedr Valley, no date, quoted in Britton's R.E.S. Handbook (Volume 5, part 11) on the Scarabeoidea (1956). — E. G. HANCOCK, Bolton Museum, Le Mans Crescent, Bolton, Greater Manchester.

ATOMARIA FIMETARII F. (COL.: CRYPTOPHAGIDAE) NEW TO KENT. — I was very agreeably surprised to find an example of this fine and rare *Atomaria* in my net after sweeping part of a patch of lawn and under and about some shrubs, etc., in my small back garden — in fact just behind the house — on the evening of 28th June last, shortly before dusk. It was at that time rather cool and breezy, but much of the afternoon had been warm. To date no further specimen has appeared, and as the beetle's special pabulum (*Coprinus comatus*, the "Ink-cap" fungus) was nowhere in evidence in the immediate vicinity, this individual was perhaps a straggler from some undiscovered breeding-source further afield. There would seem to be no previous Kent record of *A. fimetarii*, other than a possible ancient one given by Fowler (1889, *Col. Brit. Isl.*, 3: 332) for Dulwich — "partly in Kent" as my gazetteer informs me — which, from its antiquity, its borderline character, and the want of exact details, can in this context I think legitimately be ignored. — A. A. ALLEN, 49 Montcalm Road, Charlton, London, SE7 8QG.